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7. Author(s) Robert Davis, Phillip Harein, Ralph Heal, L.S. Henderson, Harry Incho, Charles Jackson, Robert Morgan

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ANALYSIS OF SPECIALIZED PESTICIDE PROBLEMS

INVERTEBRATE CONTROL AGENTS - EFFICACY TEST METHODS

VOLUME V

STORED PRODUCTS AND PREMISE TREATMENTS

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ENVIRONMENTAL PROTECTION AGENCY

ANALYSIS OF SPECIALIZED PESTICIDE PROBLEMS
INVERTEBRATE CONTROL AGENTS - EFFICACY TEST METHODS
VOLUME V
STORED PRODUCTS AND PREMISE TREATMENTS

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Report To The
Environmental Protection Agency

By The

American Institute of Biological Sciences
Arlington, Virginia 22209

EPA REVIEW NOTICE

This Report has been reviewed by the Office of Pesticide Programs, Criteria and Evaluation Division, and approved for publication. Approval does not signify that the contents necessarily reflect the views and policies of the Environmental Protection Agency, nor does mention of trade names or commercial products constitute endorsement of recommendation for use.

STORED PRODUCTS AND PREMISE TREATMENTS TASK GROUP

Chairman:

DR. ROBERT DAVIS
USDA-ARS Stored Product Insects
Research and Development Laboratory

DR. PHILLIP K. HAREIN
University of Minnesota

DR. RALPH E. HEAL
Oxford, Maryland

DR. L. S. HENDERSON
USDA-ARS, National Program Staff

DR. HARRY H. INCHO
FMC Corporation

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Celanese Coatings & Specialities Company

MR. ROBERT W. MORGAN
Dow Chemical Corporation

EPA Observer:
MR. ROGER PIERPONT
Criteria and Evaluation Division

AIBS Coordinators:
MR. DONALD R. BEEM
MS. PATRICIA RUSSELL

STORED PRODUCTS AND PREMISE TREATMENTS

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INTRODUCTION

This document provides a compilation of test methods that appear adequate for purposes of evaluating the effectiveness of pesticides against invertebrate pests of premises and stored products. The methods cited are not intended to exclude other valid procedures but for proprietary or other reasons are not available for reference. Similarly there is no intent to exclude new methods or improvements of current methods that may become available.

Testing should be conducted initially to determine the efficacy of a product against specific pest organisms. Once this has been established, further evidence of efficacy and usefulness of the product may require augmentation through large-scale laboratory, simulated field, or field testing procedures which closely approach actual use and which employ commercial application equipment.

The procedures presented herein primarily include methods for the invertebrate control agents considered as conventional chemical pesticides. Repellents, attractants, growth regulators, pheromones, etc., are not included unless they have been used over the years and there exists a substantial number of published results in the open literature.

GENERAL REFERENCES

Busvine, J.R. 1971. *A Critical Review of Techniques for Testing Insecticides*, 2nd ed. Commonwealth Inst. of Entomol., London. 345 pp.

Shepard, H.H., ed. 1958. *Methods of Testing Chemicals on Insects, Vol. I.* Burgess Publ. Co., Minneapolis, Minn. 356 pp.

Shepard, H.H., ed. 1960. *Methods of Testing Chemicals on Insects, Vol. II.* Burgess Publ. Co., Minneapolis, Minn. 248 pp.

GENERAL CONSIDERATIONS

The following factors should be considered in the testing and development of pesticide products for ultimate use in invertebrate control in premises and stored products: (1) the nature of the pest or pests to be controlled, (2) the pest population, (3) the uniqueness of the application site or structure, (4) the experimental design, and (5) the material or materials being treated.

The pest or pests to be controlled often cannot be distinguished as separate entities and will frequently need to be grouped according to control methods, geographical location, infested commodities or premises, life stage or form, type of damage being caused, ultimate use of an infested commodity, etc. Therefore, testing may need to be directed at the most important pest or pests, life stage of a pest complex, or at the species most resistant to a particular pesticide.

The pest population may be difficult to determine or to sample, and it will often be necessary to limit testing to the laboratory, to rear the desired pest species and artificially infest a stored commodity, or to develop a system of estimating the extent of an infestation in the field.

The uniqueness of each application site or structure and the fact that no two identical sites or structures will usually be available for field testing should be recognized and accommodated to by the researcher. Diversity will generally be the rule because of the great variety of form and volume in premises, storage and transportation facilities, commodities, manufacturing processes, building materials and surfaces to be treated, etc.

A proper experimental design is important in the development of acceptable test data and whenever possible the testing should yield results which can be statistically analyzed for significance. However, large-scale laboratory and field tests frequently cannot be conventionally designed or analyzed and may often be limited by cost of the treatment, geography, time, and manpower. Researchers not well versed in the methods of statistical analysis or biometrics may save valuable time by appropriate consultations before the testing is undertaken.

The material or materials being treated can be a variable of great magnitude. The variety of commodities a single pest can attack, the number of sites capable of harboring pests, and the types and locations of infested structures can result in a large array of possible field testing situations. The researcher should use extreme care in selecting test methods and field conditions that represent typical situations for application of the product to be registered.

REFERENCES

Finney, D.J. 1962. *Probit Analysis.* Cambridge University Press. 318 pp.

Smith, C.N., ed. 1966. *Insect Colonization and Mass Production.* Academic Press, New York and London. 618 pp.

Snedecor, G.W. 1950. *Statistical Methods,* 4th ed. The Iowa State College Press, Ames, Iowa.

Steel, G.D., and J.H. Torrie. 1960. *Principles and Procedures of Statistics.* McGraw Hill, New York.

DEFINITIONS

Aerosols are sprays dispensed in finely divided form in which 80 percent or more of the individual particles have a mean diameter of 30 microns or less and none of the particles has a diameter of more than 50 microns. The aerosol sprays may be generated by the action of propellant liquified gases or by thermal or mechanical breakup of liquid into fine droplets. The resulting particles remain suspended in the air for relatively long periods of time and serve to control flying pests upon contact.

Baits are formulations that are edible or attractive to a pest.

Coatings are formulations that are brushed, sprayed, or otherwise applied to and adhered to surfaces. They remain on the surface as a continuous film with little or no penetration.

Dusts are formulations composed of finely divided powders of organic or mineral origin which have been combined with a pesticide or which are in themselves pesticidal.

Fumigants are chemicals that are gaseous or will become gaseous used in enclosed spaces. They behave in accordance with the principles of the Gas Laws and readily distribute themselves and penetrate into cracks, crevices, and the commodity treated.

Impregnants are formulations that may be applied by pressure, injection, dipping, or other means to provide penetration of the pesticide beneath the surface and into the body of the treated object or substrate.

Micronized dusts are formulations of very finely divided powders that possess distinct flowable characteristics. They are usually dispersed into enclosed spaces with bursts of compressed gas.

Nonresidual (contact) sprays are liquids applied directly on the pest to be controlled.

Residual sprays are liquids applied as a wetting deposit on surfaces contacted by pests to produce a long-lasting biological effect.

Smokes are solid particulates which range from 0.3 to 2.0 microns in diameter which are produced by pyrogenics.

Space sprays are formulations delivered as mists or fine sprays that produce particles larger than aerosols which stay suspended in the treated space for relatively short periods of time.

Vapors are formulations that are volatilized by supplementary heat or by inherent high vapor pressure to produce a gaseous material.

PREMISE TREATMENTS

Premises are the areas within a structure, its walls, both inside and outside, and the immediate adjacent surrounding grounds. They include but are not necessarily limited to the following: residential structures; transportation facilities; commercial, industrial, and institutional buildings; food-handling establishments; animal feed lots and holding pens; farm poultry houses and yards; farm dairy structures and equipment; empty greenhouses; and empty mushroom houses.

FLYING PESTS

Flying pests usually include flies (houseflies, fruit flies, cluster flies, bottle flies, etc.), mosquitoes, wasps (mud-daubers, paper wasps, etc.), hornets, bees, flying moths, gnats, and other small flying insects. These pests may be occasional invaders of occupied structures (mosquitoes) or they may represent an infestation established within the structure (hornets, bees, cluster flies).

Since laboratory rearing and testing methods have been developed primarily for pests of medical or sanitary importance, these have become established as the standards for developing efficacy data applicable to most of the flying pests. However, many of the important flying pests on label claims are not amenable to laboratory rearing and therefore cannot be used as test organisms in the laboratory to determine product efficacy. Field testing against natural infestations of these insects will, therefore, be the only method for the product evaluation. Although field testing presents problems in experimental design, lack of standardized procedures, estimations of infestations, assessment of control efficacy, etc., it usually serves as the most practical means of assuring proper claims for registration of a product for the control of many of the flying pests.

Pesticide formulations used in the control of flying pests generally consist of products designed to kill (or repel) by direct contact (aerosols, sprays, smokes, vapors, etc.) or by residual activity (baits, sprays, wettable powders, etc.).

Aerosols are generally used as premise space treatments to control flying pests such as houseflies, cluster flies, fruit flies, mosquitoes, wasps, hornets, bees, flying moths, and other small flying insects. Laboratory testing of aerosols has presented certain problems primarily related to test chamber sizes and to the use of test insects in cages or in a free-flying state within the test chambers. Specifications of two standardized laboratory methods have been agreed upon and a number of papers have appeared in the literature outlining other possible procedures to be followed in the laboratory and field testing of aerosols.

The "Aerosol and Pressurized Space Spray Insecticides Test Method for Flying Insects," standardized as an industry test procedure by the Chemical Specialties Manufacturers Association (CSMA), represents the accepted laboratory method in the U.S. Although specifically designed for use with houseflies, it may be adapted for testing aerosols against other flying pests, such as mosquitoes.

The British Standard Institute procedure requires a large test chamber of more than 1,000 cubic feet for test aerosols against houseflies. Although this presents some advantages in controlling test dosages, the operators must be in the chambers during testing and the equipment demands a large commitment of building space. This procedure has not been adopted as an acceptable standard industry method in the U.S., although it is being used in laboratories in England and Africa.

Protocol and Methodology:

British Standard Institute. 1967. Insecticidal Efficiency of Aerosols Against Flies. B.S. 4172.

CSMA. 1971. Aerosol and pressurized space spray insecticide test method for flying insects. *Soap Chem. Spec. Blue Book* 27(4a):161-163, 191.

Space sprays are used as premise space treatments to control flying pests by direct contact. They usually consist of an active pesticide(s) suspended in a carrier such as oil, water, emulsions of oil in water, emulsions of water in oil, alcohol, etc., and are applied with handsprayers, mist or fog generators, pressurized dispensers, mechanical spray breakup devices such as, spinning disks, etc.

The laboratory testing of pest sprays has employed a large number of procedures for evaluating their effect in producing knockdown as well as kill of the test insects (usually houseflies or mosquitoes). These test procedures have been developed with free-flying as well as caged insects in large test chambers, small test chambers, wind tunnels, settling mist towers, etc.

The Peet-Grady Method, developed as a standardized industry test procedure by CSMA, is generally accepted for evaluating the effectiveness of space sprays under laboratory conditions. Although the housefly is specified in the procedures, it may be adapted for use against other flying insects, such as mosquitoes.

An alternative laboratory method has been devised for testing the effect of sprays against houseflies and mosquitoes at the USDA-ARS, Insects Affecting Man Research Laboratory and at some other laboratories. The procedure uses a wind tunnel to draw the spray through cages of insects (see Exhibit 1).

Protocol and Methodology:

CSMA. 1971. The Peet-Grady Method. *Soap Chem. Spec. Blue Book* 47 (4a): 158-160.

USDA-ARS. Tests with contact insecticidal aerosols against flies and mosquitoes. Insects Affecting Man Research Laboratory, Gainesville, Florida. (Exhibit 1).

Smokes for use in premise treatments have not been evaluated in the U.S. by standard laboratory or field methods. However, a number of methods for bioassaying "mosquito coil"-type smokes in the laboratory have been published.

Protocol and Methodology:

Fales, J.H., G.D. Mills, and C.G. Dubin, Jr. 1963. Evaluation of smoke from insecticidal coils against mosquitoes. *Mosq. News* 28(4): 547-553.

Mace, E.F. 1969. Biological test method for mosquito coils. *Pyrethrum Post* 10(1): 41-43.

Maciver, D.R. 1964. Mosquito coils. Part II. Studies on the action of mosquito coil smokes on mosquitoes. *Pyrethrum Post* 7(3): 7-17.

Vapors are used to control flying insects in enclosed spaces. Control is achieved by very small amounts of the insecticides, therefore, smallscale laboratory testing has been limited to the evaluation of carefully measured quantities of the vaporizing material in a solution. Tests of the actual vaporizing solid products have been developed in room-size test chambers using houseflies and mosquitoes as test insects.

Protocol and Methodology:

Batth, S.S., J. Singh, and D.C. Villeneuve. 1973. Dichlorvos vaporizers: Method for evaluating under simulated household uses. *J. Econ. Entomol.* 66(1): 146-150.

Batth, S.S., and J. Singh. 1974. Evaluation of dichlorvos vaporizing solids for controlling insects. *Can. Entomol.* 106(1): 31-37.

Khattat, F.H., and J.R. Busvine. 1965. A modified test method for measuring resistance to dichlorvos vapours. *Bull. Wld. Hlth. Org.* 32: 551-556.

Maddock, D.R. 1961. Dosage-mortality response of *Musca domestica* exposed to DDVP vapour. *Bull. Wld. Hlth. Org.* 24: 643-644.

Micronized dusts are insecticidal dusts in a very finely divided form which have been found to be effective in the direct contact control of flying insects when dispersed in enclosed areas such as trucks, aircraft, warehouses, etc. The following references are provided for possible use in the testing of micronized dusts.

Protocol and Methodology:

Jakob, W.L., D.K. Maddock, H.F. Schoof, and J.E. porter. 1972. Gas-propelled aerosols and micronized dusts for the control of insects in aircraft. 5. Effectiveness against insects of public health importance. *J. Econ. Entomol.* 65(5): 1454-1458.

Schechter, M.S., and W.N. Sullivan. 1972. Gas-propelled aerosols and micronized dusts for the control of insects in aircraft. 2. Pesticide formulations. *J. Econ. Entomol.* 65(5): 1444-1447.

Steiner, L.F., and F. Lopez-D., and J.R. Woodley. 1972. Gas-propelled aerosols and micronized dusts for the control of insects in aircraft. 3. Effectiveness against free flying Caribbean fruit flies. *J. Econ. Entomol.* 65(5): 1447-1450.

Sullivan, W.N., M.S. Schechter, C.M. Amyx, and E.E. Crooks. 1972. Gas-propelled aerosols and micronized dusts for the control of insects in aircraft. 1. Test protocol. *J. Econ. Entomol.* 65(5): 1442-1444.

Residual sprays are used to apply residual deposits of pesticides for the control of flying pests in a multitude of situations. Housefly control is obtained in and around dairy barns by the application of residual oil or water-based sprays, wettable powders, emulsions, etc. Mosquitoes are controlled by residual sprays in structures with natural infestations, or where resting may occur. Spray treatments of wasps' or hornets' nests will provide residues toxic to the returning adult insects as well as control of the larvae.

Testing of residual sprays has not been standardized in the laboratory or in field applications, however, literature is available in which satisfactory methods are outlined for possible use in obtaining efficacy data. The laboratory procedures generally consist of treatments applied to a representative surface followed by periodic testing with houseflies or mosquitoes until the residue is no longer effective. The field testing is usually conducted in infested structures (such as dairy barns) with pretreatment and post-treatment counts made of adult resting insects. In most cases the efficacy is assessed by the relative activity of the residual product in controlling flying insects over a long period of time.

Protocol and Methodology:

Anonymous. 1963. Insecticide resistance and vector control. Annex 15A.

Instructions for the bioassay of insecticidal deposits on wall surfaces. *WHO Tech. Rep. Ser.* 1963. p. 265.

Bailey D.L., G.C. LaBrecque, and T.L. Whitfield. 1970. Insecticides applied as low-volume and conventional sprays to control larvae of the house fly in poultry houses. *J. Econ. Entomol.* 63(3): 891-893.

Batth, S.S. 1974. A method recommended for evaluating residual pesticides for house fly control. *Can. Entomol.* 106(12): 1241-1246.

Brady, U.E., Jr., D.W. Meifert, and G.C. LaBrecque. 1966. Residual sprays for the control of house flies in field tests. *J. Econ. Entomol.* 59(6): 1522-1523.

Darwazeh, H.A. 1972. Preliminary evaluation of simplified technique for insecticide bioassay of adult mosquitoes. *Calif. Vector Views* 19(a): 65-66.

Fay, R.W., and D.A. Lindquist. 1954. Laboratory studies on factors influencing the efficiency of insecticide impregnated cords for house fly control. *J. Econ. Entomol.* 47(6): 975-980.

Flynn, A.D., and H.F. Schoof. 1966. Effect of surface on residual activity of selected compounds. *J. Econ. Entomol.* 59(3): 678-681.

LaBrecque, G.C., J.B. Gahan, and D.E. Weidhaas. 1971. Evaluation of various insecticides as residual sprays in buildings naturally infested with *Anopheles quadrimaculatus*. *Mosq. News* 31(2): 206-208.

Kilpatrick, J.W., and H.F. Schoof. 1963. Adult house fly control with residual treatments of six organophosphorous compounds. *J. Econ. Entomol.* 56(1): 79-81.

Wilson, H.G., G.C. LaBrecque, and J.A. Thomas. 1974. New insecticides that show residual toxicity to *Anopheles quadrimaculatus* Say. *Mosq. News* 34(1): 121-122.

USDA-ARS. Residual sprays against houseflies and/or mosquitoes. Insects Affecting Man Research Laboratory, Gainesville, Florida. (Exhibit 2).

Baits containing pesticides serve to control flying insect pests, primarily houseflies in dairy barns.

The following selected references contain descriptions of satisfactory methods for testing baits to proved efficacy data.

Protocol and Methodology:

Bailey, D.L., G.C. LaBrecque, D.W. Meifert, and P.M. Bishop. 1968.

Instructions for the bioassay of insecticidal deposits on wall surfaces. *WHO Tech. Rep. Ser.* 1963. p. 265.

Bailey D.L., G.C. LaBrecque, and T.L. Whitfield. 1970. Insecticides applied as low-volume and conventional sprays to control larvae of the house fly in poultry houses. *J. Econ. Entomol.* 63(3): 891-893.

Batth, S.S. 1974. A method recommended for evaluating residual pesticides for house fly control. *Can. Entomol.* 106(12): 1241-1246.

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Darwazeh, H.A. 1972. Preliminary evaluation of simplified technique for insecticide bioassay of adult mosquitoes. *Calif. Vector Views* 19(a): 65-66.

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Protocol and Methodology:

Bailey, D.L., G.C. LaBrecque, D.W. Meifert, and P.M. Bishop. 1968.

Insecticides in dry sugar baits against two strains of house flies. *J. Econ. Entomol.* 61(3): 743-747.

Kilpatrick, J.W., and H.F. Schoof. 1959. A semiautomatic liquid fly bait dispenser. *J. Econ. Entomol.* 52(4): 775-776.

LaBrecque, G.C., H.G. Wilson, and J.B. Gahan. 1959. Synergized pyrethrins and allethrin baits for the control of resistant house flies. *J. Econ. Entomol.* 51(6): 798-800.

CRAWLING PESTS

Crawling invertebrate pests of premises include a diverse group of insect and other arthropod species that commonly inhabit or may become occasional residents or casual invaders of structures. The principal pests encompassed by this group include residents such as: cockroaches, silverfish, firebrats, spiders, carpet beetles, fleas, bedbugs, booklice and psocids; and invaders such as: ants, clover mites, ticks (principally brown dog tick), crickets, earwigs, sowbugs (pillbugs), centipedes, millipedes, boxelder bugs, springtails (Collembola), and scorpions. Control of these pests may be important for any of several reasons including sanitation (health), damage (to materials that constitute sources of food or habitation), annoyance (nuisance). or offensiveness.

Crawling pests occur or may be found in numerous locations within or around premises depending on the habits of the individual pest involved. Typical sites include dark corners of rooms and closets; cracks and crevices in walls and between different elements of construction; along and behind baseboards; places of entrance such as around doors and windows; beneath and behind sinks, appliances, cabinets, and other fixtures and equipment; under floor coverings and furniture; around plumbing and other utility installations; pet beds and resting quarters; in and around floor drains, etc. It is to these areas and surfaces that residually active deposits of pesticides are commonly applied for control purposes. Supplementary control may be achieved by the use of nonresidual (contact) sprays, space sprays, and baits. Where pest populations are unusually large or are so firmly established that use of a residual pesticide would be unlikely to result in the immediate control desired fumigants, aerosols, mist sprays, or similar methods of control may also be employed.

Evaluation of pesticides for control of crawling pests may require both laboratory and field tests. The former procedures often allow factors such as minimum effective dosage, speed (knockdown) and duration (residuality) of effectiveness, and the effect of different surface types on biological activity to be determined, whereas field testing is usually necessary to confirm the validity of these factors under use conditions and in accordance with proposed labeling. The following references cite test procedures that have been shown to be useful.

Protocol and Methodology:

Anonymous. 1974. Insects, resistance of textiles to. AATCC Test Method 24-1975. *Am. Assoc. Tex. Color Chem.* Technical Manual pp. 263-267.

Batth, S.S. 1974. A method recommended for evaluating residual pesticides for cockroach control. *Can. Entomol.* 106(10): 1081-1085.

Bennett, G.W., and L.K. Antons. 1975. Controlling German cockroaches with pyrethrin ULV applications. *Pest Control* 43(1): 24, 26, 28.

Burden, G.S., and B.J. Smittle. 1968. Laboratory methods for evaluation of toxicants for the bedbug and the Oriental rat flea. *J. Econ. Entomol.* 61(6): 1565-1566.

Burden, G.S., and B.J. Smittle. 1969. Baygon® in field tests against German cockroaches. *J. Econ. Entomol.* 62(1): 262-263.

Burden, G.S., W.A. Banks, and E.E. Madden. 1972. Chlorpyrifos (Dursban) in field tests against German cockroaches. *Pest Control* 40(3): 13-14.

Burden, G.S. 1975. Repellency of selected insecticides. *Pest Control* 43(6): 16-18.

Burden, G.S., and E.E. Madden. 1975. *Periplaneta americana* - comparative susceptibility to residuals. *Pest Control* 43(1): 20.

CSMA. 1971. Cockroach aerosol test method. *Soap Chem. Spec. Blue Book* 47(4a): 164-165, 191.

CSMA. 1971. Cockroach spray method. *Soap Chem. Spec. Blue Book* 47(4a): 166-167.

CSMA. 1971. Textile resistance test. *Soap Chem. Spec. Blue Book* 47(4a): 168-171.

Fales, J.H., and O.F. Bodenstein. 1963. How to field test for cockroach susceptibility to chlordane. *Pest Control* 31: 18, 20, 22, 62.

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Flynn, A.D., and H.F. Schoof. 1966. Effect of surface on residual activity of selected compounds. *J. Econ. Entomol.* 59(3): 678-681.

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Goodhue, L.D. 1960. New techniques for screening cockroach repellents. *J. Econ. Entomol.* 53(5): 805-810.

Grayson, J.M. 1975. Cockroach control research in 1974. *Pest Control* 43(4): 17-20.

Hagstrum, D.M. 1970. Laboratory studies on the effect of several insecticides on *Tarentula kochi*. *J. Econ. Entomol.* 63(6): 1844-1847.

Moore, R.C. 1973. Cockroach proofing. *Conn. Agr. Exp. Sta. Bull.* New Haven 740: 1-13.

Nelson, V.A. 1969. Durshan® for control of the brown dog tick. *J. Econ. Entomol.* 62(3): 719-720.

Norment, B.R., and T.L. Pate. 1968. Residual activity of diazinon and lindane for control of *Loxosceles reclusa*. *J. Econ. Entomol.* 61(2): 574-575.

Reierson, D.A. 1973. Field tests to control German cockroaches with ULV aerosol generators. *Pest Control* 41(1): 26, 28, 31, 32.

Sterling, H.R., R.G. Price, and K.O. Furr. 1972. Laboratory evaluation of insecticides on various surfaces and at various intervals for control of the brown recluse spider. *J. Econ. Entomol.* 65(4): 1071-1073.

USDA-ARS. Insecticide residue tests with the German cockroach, *Blattella germanica* (L.). Insects Affecting Man Research Laboratory, Gainesville, Florida. (Exhibit 3).

USDA-ARS. Insecticide residue tests with the bedbug, *Cimex lectularius* (L.). Insects Affecting Man Research Laboratory, Gainesville, Florida. (Exhibit 4).

STORED-PRODUCT TREATMENTS

Invertebrate pests of stored products include both the insects and mites that damage and contaminate harvested raw agricultural commodities and subsequent processed or manufactured products. The subject pests must be controlled not only in the commodities but in facilities where they are stored, processed, manufactured, packaged, and transported.

The majority of the primary insects attacking food, feed, and seed are beetles or moths. Most species are cosmopolitan in distribution and at least 25 are economically important pests of stored products in the United States. Some species cause damage in both the larval and adult stages.

General References

Boles, H.P., and F.O. Marzke. 1966. Lepidoptera infesting stored products. Pages 259-270 in C.N. Smith, ed. *Insect Colonization and Mass Production.* Academic Press, New York and London. 618 pp.

Harein, P.K., and E. De Las Casas. 1974. Chemical control of stored-grain insects and associated micro- and macro-organisms. Pages 232-291 in *Storage of Cereal Grains and Their Products.* American Association of Cereal Chemists. St. Paul, Minn.

Harein, P.K., and E.L. Soderstrom. 1966. Coleoptera infesting stored products. Pages 251-257 in C.N. Smith, ed. *Insect Colonization and Mass Production.* Academic Press, New York and London. 618 pp.

Aerosol applications for stored-product insects are effective in closed and semiclosed areas such as warehouses, transporting vehicles, and various processing and manufacturing areas. These treatments are effective to prevent or protect against infestations but have little potential control of established infestations.

Protocol and Methodology:

CSMA. 1971. Aerosol and pressurized space spray insecticide test method for flying insects. *Soap Chem. Spec. Blue Book* 47(4a): 161-163, 191.

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Cogburn, R.R., and R.A. Simonaitis. 1975. Dichlorvos for control of stored-product insects in port warehouses. Low volume aerosols and commodity residues. *J. Econ. Entomol.* 68(3): 361-365.

Gillenwater, H.B., G. Eason, and E.B. Bauman. 1972. Gas-propelled aerosols and micronized dusts for control of insects in aircraft. 4. A potential for controlling stored-product insects. *J. Econ. Entomol.* 65(5): 1450-1453.

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Schechter, M.S., and W.N. Sullivan. 1972. Gas-propelled aerosols and micronized dusts for control of insects in aircraft. 2. Pesticide formulations. *J. Econ. Entomol.* 65(5): 1444-1447.

Steiner, L.F., F. Lopez-D., and J.R. Woodley. 1972. Gas-propelled aerosols and micronized dusts for control of insects in aircraft. 3. Effectiveness against free flying Caribbean fruit flies. *J. Econ. Entomol.* 65(5):1447-1450.

Sullivan, W.N., M.S. Schechter, C.M. Amyx, and E.E. Crooks. 1972. Gas-propelled aerosols and micronized dusts for control of insects in aircraft. 1. Test protocol. *J. Econ. Entomol.* 65(5): 1442-1444.

Yerington, A.P. 1967. Control of Drosophila in wineries with dichlorvos aerosols. *J. Econ. Entomol.* 60(3): 701-704.

Fumigants must reach the insect as a gas to be effective whether dispersed as a solid, liquid, or gas, in lethal concentrations for practical exposure periods. To maintain such concentrations the product, equipment, or area treated must be relatively gas-tight. This is generally attained by sealing the structure, enclosing the infested items under gas-tight tarpulins, or placing it in a fumigation chamber. Fumigants have no residual effectiveness.

Protocol and Methodology:

Childs, D.P. 1967. Cigarette beetle control in warehouses with HCN and dichlorvos. *J. Econ. Entomol.* 60(1): 263-265.

Cogburn, R.R. 1974. Detia Ex-B® for phosphine fumigation in sacked milled rice. *J. Econ. Entomol.* 67(3): 436-438.

Cogburn, R.R., and E.W. Tilton. 1963. Studies of phosphine as a fumigant for sacked rice under gas-tight tarpaulins. *J. Econ. Entomol.* 56(5): 706-708.

Cooper, C.V., and H.B. Gillenwater. 1972. Preliminary evaluation of six candidate fumigants against stored-product insects. *J. Ga. Entomol. Soc.* 7(4): 250-253.

Cooper, C.V., J.W. Press, and H.B. Gillenwater. 1970. The fumigant potential of GC-10033 against stored-product insects. *J. Econ. Entomol.* 63(6): 1979-1981.

Howe, R.W. 1974. Problems in the laboratory investigation of the toxicity of phosphine to stored product insects. *J. Stored Prod. Res.* 10(3/4): 167-181.

Kirkpatrick, R.L. 1966. Toxicity of seven candidate fumigants to stored-product insects. *J. Econ. Entomol.* 59(3): 558-560.

Leesch, J.G., H.B. Gillenwater, and J.O. Woodward. 1974. Methyl bromide fumigation of shelled peanuts in bulk containers. *J. Econ. Entomol.* 67(6) 769-771.

Lindgren, D.L., and L.E. Vincent. 1966. Relative toxicity of hydrogen phosphide to various stored-product insects. *J. Stored Prod. Res.* 2(2): 141-146.

Monro, H.A.U., C.R. Cunningham, and J.E. King. 1952. Hydrogen cyanide and methyl bromide as fumigants for insect control in empty cargo ships. *Sci. Agric.* 32: 241-265.

Soles, R.L., and P.K. Harein. 1962. The fumigant toxicity of two new chemicals to stored-product insects. *J. Econ. Entomol.* 55(6): 1014-1015.

Storey, C.L. 1967. Comparative Study of Methods of Distributing Methyl Bromide in Flat Storages of Wheat: Gravity-penetration, Single Pass, and Closed-recirculation. USDA-ARS Mktg. Res. Rept. No. 794. 16 pp.

Storey, C.L., J.K. Quinlan, and L.T. Davidson. 1970. Distribution and Retention to Fumigant Components in Shelled Corn in 3,250 Bushel Metal Bins. USDA-ARS Mktg. Res. Rept. No. 897. 12 pp.

Tilton, E.W., and R.R. Cogburn. 1965. Phosphine fumigation of rough rice in upright bins. *Rice J.* 68(11): 8-9.

USDA-ARS. Penetration and toxicity of fumigants for potential use against stored-product insects. Stored-Product Insects Research and Development Laboratory, Savannah, Georgia. (Exhibit 5).

Vardell, H.H., A. Cagle, and E. Cooper. 1973. Fumigation with phosphine against stored-product insects in bagged flour in plywood overpacks. *J. Econ. Entomol.* 66(5): 1209-1210.

White, G.D., H.H. Walkden, and H.D. Nelson. 1967. Evaluation of three spot fumigant formulations. *Milling Prod.* 5(22): 15-20.

Insect-Resistant Packaging (protective containers) for food and feeds have found only limited application in the marketing channels. The availability of pesticides with low mammalian toxicity and the economics of treating food packages have been the limiting factors. However, there are several approved uses of insect-resistant packaging, and there are several adequate examples of laboratory and simulated and/or actual field procedures presented in the literature.

Insect-exposure rooms for conducting the tests should be maintained at temperatures and relative humidities favorable for insect development. Temperature and relative humidity should be determined periodically throughout the test. Insect populations should be kept active so that the test packages are under constant attack by the test insects. Insect species in the exposure room should include as a minimum those species that are particularly troublesome to the packaged commodities being tested. The room should also contain the species commonly found in food storage situations, such as red flour beetle *Tribolium castaneum* or confused flour beetle *T. confusum*, lesser grain beetle *Rhyzopertha dominica*, cigarette beetle *Lasioderma serricorne*, sawtoothed grain beetle *Oryzaephilus surinamensis* or merchant grain beetle *O. mercator*, Indian meal moth *Plodia interpunctella*, and *Trogoderma variable*. Approxpriate insect food media should be distributed throughout the room as a supplementary food source. It may be desirable or necessary to place additional insect cultures in the room from time to time if the population of a given species decreases noticeably.

The packages should be arranged in the room to provide maximum exposure to the insects. However, the packages must be arranged in such a manner that would simulate a realistic situation. For example, if the repellent-treated packages are usually contained in external packages in normal marketing channels, the test packages should be in a similar external container for exposure to insects and determination of repellent residues in the packaged product.

As a minimum, the test variables will include the experimental (repellent-treated package), an untreated control (same package construction but with no treatment), and a check package (same packaging material but with seals that would allow insects to enter easily). The untreated control demonstrates the necessity for the repellent treatment, while the check package demonstrates that (1) the commodity is attacked by the insects and (2) the insects are sufficiently active to provide a reliable test.

Representative packages will be examined for insect penetration and for insects in the commodity at frequent intervals throughout the test. The package construction such as seals, closure, glue lines, and heat-sealed areas should be examined for openings that would allow insects to enter the packages. The test storage period will extend for at least the maximum storage period encountered by the package and product being tested.

Protocol and Methodology:

Davis, D.F., and H. Laudani. 1956. Long-term insecticide tests. *Modern Packaging* 29(7): 236-240, 332, 334, 337, 338.

Gillenwater, H.B., and L.L. McDonald. 1975. Repellency of nineteen compounds to adult *Tribolium confusum*. *J. Ga. Entomol. Soc.* 10(2): 151-155.

Guy, R.H., H.A. Highland, and C.E. Metts. 1970. Repellency of selected compounds to *Tribolium confusum*. *J. Econ. Entomol.* 63(6): 1847-1850.

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Highland, H.A., E. Mangum, Jr., and W.H. Schoenherr. 1970. Values of insect-resistant cotton bags in overseas shipments examined. *Northwestern Miller* 277(3): 8-12.

Highland, H.A., and P.H. Merritt. 1973. Synthetic pyrethroids as package treatments to prevent insect penetration. *J. Econ. Entomol.* 66(2): 540-541.

Incho. H.H., E.J. Incho, and N.W. Matthews. 1953. Insect-proofing paper. *Agr. Food Chem.* 1(20): 1200-1203.

Laudani, H., D.F. Davis, and G.R. Swank. 1955. A laboratory method of evaluating the repellency of treated paper to stored-product insects. *Tech. Assoc. Pulp Paper Ind.* 38(6): 336-341.

Laudani, H., H.A. Highland, and E.G. Jay. 1966. Treated bags keep cornmeal insect-free during overseas shipment. *Am. Miller Process.* 94(2): 14-19, 33.

Loschiavo, S.R. 1961. 4'(3,3-dimethyl-1-triazeno) acetanilide to protect package cereals against stored products insects. *Food Tech.* 24(4): 181-185.

USDA-ARS. Preliminary evaluation of new candidate materials as repellents to stored-product insects. Stored-Product Insects Research and Development Laboratory, Savannah, Georgia. (Exhibit 6).

Yerington, A.P. 1971. Insect resistance of dried-fruit packages. *Mod. Packag.* 44(6): 76, 77, 80.

Residual protectants are applied to grains or oil seeds during the postharvest period usually as they are going into their first major storage period. However, residual protectants may be applied at other times during the storage period prior to processing. The objective of this protective treatment is to control insect pests upon emergence from the commodity and to prevent spread of insect pest infestation of the commodity during the storage period. Surface treatments of a residual protectant may also be applied during the storage period to prevent reinfestation. Residual protectants are usually formulated as sprays such as water-wettable powders and emulsions or dust.

Efficacy of a residual protectant may be determined satisfactorily by any one of several laboratory procedures. Usefulness may be determined by either simulated field tests or pilot-scale field tests.

Protocol and Methodology:

Cogburn, R.R. 1967. Laboratory tests of five new insecticides as protectants for stored rough rice. *J. Econ. Entomol.* 60(5): 1286-1289.

Harein, P.K., and H.B. Gillenwater. 1966. Exploratory tests with bromodan as a protectant for wheat against stored-product insects. *J. Econ. Entomol.* 59(2): 413-414.

Kirkpatrick, R.L., P.K. Harein, and C.V. Cooper. 1968. Laboratory tests with dichlorvos applied as a wheat protectant against rice weevils. *J. Econ. Entomol.* 61(2): 356-358.

La Hue, D.W. 1966. Evaluation of Malathion, Synergized Pyrethrum, and Diatomaceous Earth on Shelled Corn as Protectants Against Insects in Small Bins. USDA-ARS Mktg. Res. Rpt. No. 768. 10 pp.

La Hue, D.W. 1967. Evaluation of Malathion, Synergized Pyrethrum, and a Diatomaceous Earth as Protectants Against Insects in Sorghum Grain in Small Bins. USDA-ARS Mktg. Res. Rpt. No. 781. 11 pp.

La Hue, D.W. 1973. Gardona as a protectant against insects in stored wheat. *J. Econ. Entomol.* 66(2): 485-489.

La Hue, D.W., and E.B. Dicke. 1971. Phoxim as an insect protectant for stored grain. *J. Econ. Entomol.* 64(4): 1530-1533.

La Hue, D.W., and C.C. Fifield. 1967. Evaluation of Four Inert Dusts on Wheat as Protectants Against Insects in Small Bins. USDA-ARS Mktg. Res. Rpt. No. 780. 24 pp.

Laudani, H., H.B. Gillenwater, G.H. Kantack, and M.F. Phillips. 1959. Protection of citrus pulp against insect infestation with surface applications of pyrethrum-piperonyl butoxide wettable powder. *J. Econ. Entomol.* 52(2): 224-227.

Nelson, H.D., G.H. Spitler, and A.P. Yerington. 1967. Use of Malathion-treated Drying Trays to Protect Raisins from Insects During Drying and Storage. USDA-ARS Mktg. Res. Rpt. No. 789. 14 pp.

Phillips, G.L. 1959. Control of insects with pyrethrum sprays in wheat stored in ship's holds. *J. Econ. Entomol.* 52(4): 557-559.

Press, J.W., H.B. Gillenwater, and G. Eason. 1970. Dichlorvos treatments to prevent dissemination of the white-fringed beetle, *Graphognathus leucoloma*, in shipped wheat (Coleoptera: Curculionidae). *J. Ga. Entomol. Soc.* 5(3): 158-162.

Spitler, G.H., P.L. Hartsell, and H.D. Nelson. 1974. Malathion Protection of Inshell Almonds in Bulk Storage--Pilot Scale Study. ARS W-26. 8 pp.

Strong, R.G. 1970. Relative susceptibility of confused and red flour beetles to 12 organophosphorous insecticides with notes on adequacy of the test method. *J. Econ. Entomol.* 63(1): 258-263.

Strong, R.G., and D.E. Sbur. 1961. Evaluation of insecticides as protectants against pests of stored grain and seeds. *J. Econ. Entomol.* 54(2): 235-238.

Strong, R.G., and D.E. Sbur. 1964. Protective sprays against internal infestation of grain beetles in wheat. *J. Econ. Entomol.* 57(4): 544-548.

Tilton, E.W., and R.R. Cogburn. 1967. Laboratory evaluation of fenthion for the protection of rough rice against insect attack. *J. Econ. Entomol.* 60(1): 233-235.

USDA-ARS. Test method for initial evaluation of promising insecticides as protectants for commodities. Stored-Product Insects Research and Development Laboratory, Savannah, Georgia. (Exhibit 7).

Vardell, H.H., H.B. Gillenwater, G. Eason, and A. Cagle. 1973. White-fringed beetles: Dichlorvos applied as a postharvest treatment to protect wheat. *J. Econ. Entomol.* 66(1): 225-227.

Residual sprays are commonly applied on surfaces where pests alight, crawl, hide, feed, or reproduce. Common examples are found in warehouses, in mills and processing establishments, in grain bins, and in transportation facilities.

Protocol and Methodology:

Harein, P.K., and J.H. Schesser. 1975. A Gardona-Vapona mixture for control of stored-product insects in railway cars. *J. Econ. Entomol.* 68(1):113-118.

McDonald, L.L., R.H. Guy, and R.D. Speirs. 1970. Preliminary Evaluation of New Candidate Materials as Toxicants, Repellents, and Attractants Against Stored-product Insects. I. USDA-ARS Mktg. Res. Rpt. No. 882. 8 pp.

Speirs, R.D., and J.H. Lang. 1970. Contact, Residue, and Vapor Toxicity of New Insecticides to Stored-product Insects. II. USDA-ARS Mktg. Res. Rpt. No. 885. 35 pp.

USDA-ARS. Test for residual toxicity against stored-product insects. Stored-Product Insects Research and Development Laboratory, Savannah, Georgia. (Exhibit 8).

Vapors are useful in situations similar to those of aerosols and space sprays. They are not known to penetrate commodities in concentrations lethal to the target pest.

Protocol and Methodology:

Batth, S.S., J. Singh, and D.C. Villaneuve. 1973. Dichlorvos vaporizers. Method for evaluation under simulated household use. *J. Econ. Entomol.* 66(1): 146-150.

Gillenwater, H.B., P.K. Harein, E.W. Loy, Jr., J.F. Thompson, H. Laudani, and G. Eason. 1971. Dichlorvos applied as a vapor in a warehouse containing packaged foods. *J. Stored Prod. Res.* 7(1): 45-56.

Harein, P.K., H.B. Gillenwater, and G. Eason. 1971. Dichlorvos space treatments for protection of packaged flour against insect infestation. *J. Stored Prod. Res.* 7(1): 57-62.

Harein, P.K., H.B. Gillenwater, and E.G. Jay. 1970. Dichlorvos: Methods of dispensing, estimates of concentrations in air, and toxicity to stored-product insects. *J. Econ. Entomol.* 63(4): 1263-1264.

Speirs, R.D., and J.H. Lang. 1970. Contact, Residue, and Vapor Toxicity of New Insecticides to Stored-product Insects. II. USDA-ARS Mktg. Res. Rpt. No. 885. 35 pp.

STRUCTURES AND STRUCTURAL MATERIAL TREATMENTS (TERRESTRIAL)

Wood-destroying beetles include species of Anobiidae, Lyctidae, Bostrichidae, and one species of Cerambycidae, *Hylotrupes bajulus* L. (the old house borer), all of which may reinfest structural members or lumber in storage. Occasionally other species may infest dead or dying trees or drying lumber and emerge after the wood has been placed in use, but are incapable of reinfesting the wood. Examples are long-horned borers, flatheaded borers, ambrosia beetles, and pinworms. The use of pesticides to control these non-reinfesting species is not needed except where the infestation is extensive.

Control of wood-destroying beetles in structural members or in stored lumber may be by (1) residual spray or impregnation with a pesticide or (2) fumigation. Prevention of attack by wood-destroying beetles may be achieved through residual spray or impregnation. There are no standard tests recognized in the United States for the laboratory evaluation of residual sprays or impregnants in the prevention or elimination of infestations by these species. No fumigation tests have been specified for these species but methods parallel to those used for stored products may be utilized.

Protocol and Methodology:

Becker, W.B., H.G. Abbott, and J.H. Rich. 1956. Effect of lindane emulsion sprays on the insect invasion of white pine sawlogs and the grade yield of the resulting lumber. *J. Econ. Entomol.* 49(5): 664-666.

Wharf borer adults emerging into buildings or in areas around the sites of former buildings may be controlled by treatments designed for the control of flying insects; and these could be evaluated by tests paralleling those for other household pests.

Carpenter ant and carpenter bee controls have been evaluated chiefly through the observation of the effect of their applications under field conditions.

Subterranean termite attacks in a structure may be prevented or controlled through impregnation with a termite toxicant of the soil beneath and adjacent to the structure. The effectiveness of such treatments is measured by the time over which the toxic barrier remains effective in resisting penetration by the termites. No published laboratory methods are currently recognized as giving a reliable evaluation of soil toxicants for termite control.

Three field tests are recognized as giving a reliable evaluation of soil toxicants under conditions paralleling rigorous exposure in practical application. These tests are known as: (1) The Stake Method

(Exhibit 9), (2) The Ground Board Method (Exhibit 10), and (3) The Modified Ground Board Method (Exhibit 11). These methods were developed and validated at the Southern Forest Experiment Station of the U.S. Department of Agriculture. Interpretation of these tests has been that effectiveness retained over a period of 5 years is acceptable evidence of efficacy.

Protection of wood from attack by subterranean termites may also be provided by impregnation of the wood by a termite toxicant. Such treatments may be evaluated by a laboratory method developed by the American Wood Preservers Association (Exhibit 12).

Protocol and Methodology:

American Wood Preservers Association. Standard Method for Laboratory Evaluation to Determine Resistance to Subterranean Termites. AWPA, Washington, D.C. Standard M12-70. (Exhibit 12).

Johnson, H.R., V.K. Smith, and R.H. Beal. 1971. Chemicals for subterranean termite control: Results of long-term tests. *J. Econ. Entomol.* 64(3): 745-748.

USDA-Forest Service. Standard Stake Method. This method was provided by and is currently in use at the Wood Products Insect Laboratory, Southern Forest Experiment Station, Gulfport, Miss. (Exhibit 9).

USDA-Forest Service. Standard Ground-Board Method. This method was provided by and is currently in use at the Wood Products Insect Laboratory, Southern Forest Experiment Station, Gulfport, Miss. (Exhibit 10).

USDA-Forest Service. Modified Ground-Board Method. This method was provided by and is currently in use at the Wood Products Insect Laboratory, Southern Forest Experiment Station, Gulfport, Miss. (Exhibit 11).

Damp wood: Efficacy of materials against the subterranean termite has been the practical guide to the effectiveness of a pesticide for these pests. No laboratory or field tests for their evaluation are in the literature.

Dry wood: These termites in a structure or its furnishings are controlled by fumigation or treatment of channels in the wood with pesticidal dusts or liquids. Evaluation of the treatments of channels with dusts, liquids, or spot fumigants for the elimination of drywood termites has been chiefly through the observation of the effect of such treatments under field conditions. Absorptive dusts have been used as a protective treatment and likewise have been evaluated on the basis of field treatments.

Protocol and Methodology:

Bess. H.A., and A.K. Ota. 1960. Fumigation of buildings to control the dry-wood termite, *Cryptotermes brevis*. *J. Econ. Entomol.* 53(4): 503-510.

Stewart, D. 1957. Sulfuryl fluoride - a new fumigant for control of the drywood termite, *Kalotermes minor* Hagen. *J. Econ. Entomol.* 50(1): 7-11.

STRUCTURES AND STRUCTURAL MATERIAL TREATMENTS (MARINE)

Boring pests. Teredos and Limnoria are the important borers which cause damage to wooden marine structures. Pesticides formulated as impregnants and surface coatings are used to protect marine structures against borer attack.

No standard test method has been established for laboratory testing samples, either treated with impregnants or painted with surface coatings, for protection against borers. Tests under actual marine exposure conditions where borers naturally occur should be made. All tests must include untreated blank samples of the same material as the product being treated and preferably samples treated with a product of known and acceptable performance. Evaluation of the effectiveness of the proposed treatment is based on a comparison of the attack of the borers on the treated versus the untreated specimens. Exposures should be of two types: (a) waterline and (b) completely submerged; products for use on pilings and other wooden marine support structures should also be exposed to include a mud-line area. Enough replicates should be exposed to satisfactorily substantiate the effectiveness of the treatment under test. Examination and evaluation should be made at intervals sufficient to determine the progression of damage in the blanks and the effectiveness of the pesticide on the treated samples; total exposure time should be not less than 1 year. For the purpose of periodic examination, representative sections may be cut from the exposed test members; all sections should be cut the same size and from the same portion of the treated and untreated samples.

Marine fouling. Some of the most important marine fouling organisms are:

Algae

Annelids (tube worms)

Barnacles

Bryozoa (encrusting and branching)

Hydroids

Mulluscs

Tunicates

Pesticides in the form of surface coatings are used to control marine fouling and may be applied by brushing, spraying, or roller coating.

Although accelerated laboratory tests have been developed for testing paints containing copper as the active ingredient, they are not applicable to paints which also depend upon other active ingredients for efficacy.

The acclerated tests are used strictly for preliminary laboratory evaluation to eliminate obviously inefficient formulations from further testing.

The only practical method for determing the efficacy of an antifouling paint (coating) is to expose it under actual fouling conditions. Efficacy can be demonstrated by comparing treated (coated) panel specimens with uncoated panels, or other suitable blanks, and with panels coated with paints of known performance. Panels for such tests should be as large as possible to provide a reasonable area for examination and should not be smaller than 6 x 12 inches. The coating system should be the same in both composition (number of prime coats, when required, and antifouling coats) and application as that recommended for field application. Panels should be exposed at least in duplicate at both waterline and complete submersion, and total efficacy shall be judged on both types of exposure. Specimens should be examined at monthly intervals and efficacy determined after 12 months' exposure. Antifouling paints, for which specific lengths of time for protection against fouling is claimed, shall be exposed for not less than the time period claimed. A monthly census of fouling organisms on an untreated blank should be part of the efficacy evidence. The physical condition of the film, fouling rating compared to the untreated specimen, and overall performance should be reported for total evaluation of efficacy.

Protocol and Methodology:

Environmental Protection Agency. Tentative Recommended Practice for Testing Antifouling Paints on Wood Substrates: T5D 6.103 (Revised 1-1-75). E.P.A. Tech. Serv. Div., Chem. and Biological Investigation Branch, Beltsville, Md. 20705.

Environmental Protection Agency. Tentative Specification for Preparation of Metal Panels for Antifouling Paint Testing: T5D 6.106 (1-1-75). Tech. Serv. Div., Chem. and Biological Investigation Branch, Beltsville, Md. 20705.

Environmental Protection Agency. Tentative Specification for Standard Antifouling Test Racks: T5D 6.107 (1-1-75).

Ketchum, B.H. 1948. Action of antifouling paints. V - The use of glycine solutions as accelerated test of availability of toxicant. *Ind. Eng. Chem.* 40: 249-253.

Ketchum, B.H., J.D. Ferry, A.C. Redfield, and A.E. Burns, Jr. 1945. Evaluation of antifouling paints by leaching rate determinations. *Ind. Eng. Chem.* 37: 456-460.

Unpublished Protocol. Test method for antifouling paints on wood substrates for fresh or salt water exposure. Adapted from E.P.A. and industry sources. (Exhibit 13).

Unpublished Protocol. Test method for antifouling paints on metal substrates for fresh or salt water exposure. Adapted from E.P.A. and industry sources. (Exhibit 14).

FABRIC PROTECTIVE TREATMENTS

Fabrics are considered to be all animal fibers and blends with plant and synthetic fibers. Treatments are applied at any stages from harvest (raw) through processing and until fully utilized by man or domestic animals.

Several insects attack woolen fabric and other animal fibers; however, at least two pest species, one moth and one beetle, should be used in conducting tests for fabric protectants. Tests for efficacy and usefulness are not usually conducted as separate entities, but rather usefulness data are secured following aging and through subsequent performance tests.

Protocol and Methodology:

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USDA-ARS. Temporary fabric treatments. Stored-Product Insects Research and Development Laboratory, Savannah, GA. (Exhibit 15).

USDA-ARS. Semipermanent fabric treatments. Stored-Product Insects Research and Development Laboratory, Savannah, Ga. (Exhibit 16).

USDA-ARS. Permanent fabric treatments. ed Product Insects Research and Development Laboratory, Savannah, (Exhibit 17).

Exhibit 1

TESTS WITH CONTACT INSECTICIDAL AEROSOLS AGAINST FLIES AND MOSQUITOES

USDA-ARS, Insects Affecting Man Research Laboratory
Gainesville, Florida

A wind tunnel is used consisting of a large anterior cone tapering into a posterior cylindrical tube, 4 inches in diameter, through which a column of air is drawn at 4 m.p.h. by a suction fan. Twenty female flies, 4-5 days old, or 25 female mosquitoes, 1 to 3 days old, are confined in a 4-inch cylindrical screen cage, which is placed in the center of the tube. One-fourth ml of the pesticide usually in a deodorized kerosene solution, is atomized at 1 p.s.i. into the mouth of the cone, and the insects are exposed momentarily to the spray as it is drawn through the cage. One or two seconds after the spray has been exhausted from the tunnel, the cage is removed from the tube and the insects are anesthetized with carbon dioxide gas and transferred to clean, untreated holding cages. Sugar solution is furnished in an absorbent cotton pad. Mortality is recorded 24 hours after treatment. All tests are replicated.

The first tests are made with sprays containing 2.5 percent of the pesticide. If mortality exceeds 50 percent, lower concentrations are tested to derive a valid probit mortality-log concentration regression line.

References

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Exhibit 2

RESIDUAL SPRAYS AGAINST HOUSEFLIES AND/OR MOSQUITOES

USDA-ARS, Insects Affecting Man Research Laboratory
Gainesville, Florida

The compounds are sprayed on 12- by 12-inch plywood panels at the rate of 100 mg of active ingredient per square foot. In each test 20 female houseflies, 4 to 5 days of age, are exposed under half sections of petri dishes on the treated panels for 60 minutes. The flies are then transferred to cylindrical screen cages, provided with cotton pads saturated with sugar solution, and held for 24 hours, at which time mortality counts are made.

The panels are tested 1 week after treatment, 4 weeks after treatment, and every 4 weeks thereafter for a period of 24 weeks, or until they become ineffective, whichever occurs first. Panels are considered ineffective when they fail to produce at least 90-percent mortality. Enough panels are sprayed with each insecticide to avoid using any surface twice.

The compounds are usually tested as solutions in acetone or other volatile solvents, but occasionally wettable powders and emulsions are used, in which case the classification is based on the most effective treatment.

The same procedure for testing residual sprays against houseflies is used to determine the residual effectiveness of the compound against 1- to 2-day-old female common malaria mosquitoes, except that in this test the panels are considered ineffective when they fail to produce at least 70-percent mortality.

Classification system:

1. Ineffective at 1 week.
2. Effective for 1 week.
3. Effective for 4 weeks.
4. Effective for 8-20 weeks.
5. Effective for 24 or more weeks.

Exhibit 3

INSECTICIDE RESIDUE TESTS WITH THE GERMAN COCKROACH,
Blatiella germanica

USDA-ARS, Insects Affecting Man Research Laboratory
Gainesville, Florida

Panels, 6 by 6 inches of 1/4-inch plywood, are treated with acetone solutions of toxicants at the rate of 100 mg per square foot. When necessary, emulsions are substituted for the acetone solutions. after the treated panels are allowed to dry for 2 hours, 20 adult male cockroaches (two replicates of 10 each) are exposed to the residues for 30 minutes. Exposures to the treated surfaces are made under inverted plastic dished coated on the inner surface with pyrophyllite, which prevents the cockroaches from crawling up the sides. At the conclusion of the exposure period, the cockroaches are transferred to clean petri dishes.

Observations on mortality are made after 48 hours. The treated panels are then allowed to age and are tested at intervals of 1, 2, and 4 weeks, or longer if necessary, depending on the effectiveness of the residues.

Classification system:

1. Less than 80-percent mortality with fresh (aged 2 hours) residue.
2. 80-percent mortality with fresh residues but not after 1 week.
3. 80-percent mortality for 1 week but not 2 weeks.
4. 80-percent mortality for 2 weeks but not 4 weeks.
5. 80-percent mortality for 4 weeks or more.

Exhibit 4

INSECTICIDE RESIDUE TESTS WITH THE BEDBUG, *Cimex lectularius*

USDA-ARS, Insects Affecting Man Research Laboratory
Gainesville, Florida

Residue tests are conducted on circular pieces of Whatman No. 1 filter paper, 38 mm in diameter, impregnated with 0.2 ml of an acetone solution containing 0.31 percent of an insecticide. This impregnation rate produces a residue of 50 mg of toxicant per square foot. After drying for 1 hour, each treated paper is placed in a 50-ml beaker and ten 3-day-starved adult bedbugs are placed on it. After exposure for 24 hours, live and affected bed bugs are removed from the treated paper and placed on untreated filter paper in a clean 50-ml beaker for a holding period of 24 additional hours, after which mortality readings are made. The treated papers are aged in the beakers and tested at 1 week, 2 weeks, 4 weeks, and every 4 weeks thereafter for 24 weeks, or until less than 90-percent kill is obtained, whichever occurs first.

Classification system:

1. Less than 90-percent mortality with fresh (aged 1 hour) residue.
2. 90-percent mortality with fresh residue but not for 2 weeks.
3. 90-percent mortality for 2 weeks but not 4 weeks.
4. 90-percent mortality for 4 weeks but not 8 weeks.
5. 90-percent mortality for 8 or more weeks.

Exhibit 5

PENETRATION AND TOXICITY OF FUMIGANTS FOR POTENTIAL USE AGAINST STORED-PRODUCT INSECTS

USDA-ARS, Stored-Product Insects Research and Development Laboratory
Savannah, Georgia

For greatest practical value, a toxic gas used as a fumigant to control stored-product insects must penetrate a mass of commodity in sufficient concentrations to kill insects within the mass. A simple test of the ability of a candidate compound to penetrate a commodity and kill insects can be performed in 3.8-liter mason jars as fumitoria, using soft red winter wheat as a substrate for penetration. Any species and life stage of stored-product insects, except adults of moths, can be used as test insects.

Method

Wheat is cleaned to <1% dockage and conditioned to 12±0.1% moisture content. Mason jars (3.8 liter) equipped with standard screw-cap lids are used as fumitoria. A 5-mm diameter hole is drilled in the center of the lid, and then a paper clip to hold a piece of blotter paper is soldered to the inside of the lid near the hole. Insects are held in cages made of 40- by 36-mesh Monel wire cloth measuring 7.6 cm in length by 1.8 cm in diameter and closed on the bottom with wire and on the top with polyethylene stoppers. Twenty-five insects are placed in a cage, and then it is filled with wheat.

One kilogram of wheat is placed in the jar, and then the insect cages (one cage per species of insects being tested) are laid flat on the surface of this mass. Another kilogram of wheat is placed in the jar so that the cages are held near the center of the 2-kg mass of wheat.

Insects and grain are conditioned at the fumigation temperature in the fumitoria for 24 hours prior to testing by placing the prepared fumitoria into a controlled temperature room. After conditioning, dosing is accomplished after sealing the fumitoria, by placing the liquid compound from a microsyringe on the blotter paper through a piece of cellophane tape which covers the whole drilled in the lid directly over the paper clip. Upon removal of the syringe needle, another piece of cellophane is placed over the puncture hole. Each dosage is replicated 3 times. This method is described by Cooper et al. (1970).

If a compound in gas state is being tested; the fumitorium lid is modified by drilling a hole to accommodate a No. 5 stopper through which a piece of glass tubing extends 2-5 cm into the fumitorium. A piece of rubber tubing equipped with a pinch clamp is provided on the

outside to seal the glass tubing. Dosing is accomplished by removing slightly more air from the fumitorium than will be added as gaseous fumigant. The fumigant gas is then introduced using a syringe, and after application, the pinch clamp is released momentarily to equilibrate the negative pressure in the fumitorium to the atmospheric pressure. This method is derived from Whitney and Harein (1959).

After dosing, fumitoria are placed in a controlled temperature room for 24 hours. Upon removal, the fumitoria are aerated in a hood, and insects are removed and placed in suitable containers with food. Insects are held in a controlled temperature room during postexposure mortality observations. Final mortality observations are as follows: (1) adults of red flour beetle *Tribolium castaneum*, confused flour beetle *T. confusum* and merchant grain beetle *Oryzaephilus surinamensis* - 21 days, (2) adults of cigarette beetle *Lasioderma serricorne* - 7 days, (3) 1-month larvae of black carpet beetle *Attagenus megatoma* - 35 days. If eggs or intrakernal feeders of a species are exposed in commodity, the emergence of adults is observed until no further emergence occurs or until the normal life cycle of the insect species is complete.

Tests with new candidate compounds are always compared with data obtained in concurrent tests with a standard such as methyl bromide.

Dosage-mortality analyses are conducted on the date to obtain LD_{50}'s, LD_{90}'s, etc., and their fiducial limits. A computer program such as the probit analysis described by R.J. Daum (1970) is desirable.

References

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Exhibit 6

PRELIMINARY EVALUATION OF NEW CANDIDATE MATERIALS AS REPELLENTS TO STORED-PRODUCT INSECTS

USDA-ARS, Stored-Product Insects Research and Development Laboratory
Savannah, Georgia

The insects used in the tests are confused flour beetle adults, *Tribolium confusum* Jacquelin duVal, 7 to 14 days old. They are reared in a chamber maintained at 27°±1° C and 60±5 percent relative humidity on a diet consisting of 47.5 percent each of white flour and cornmeal and 5 percent of brewer's yeast. All testing and aging of treated papers are performed under similar conditions.

Strips of aluminum foil, laminated to 40-lb. kraft paper, 4 by 6 in., are treated on the paper side with acetone solutions of the candidate repellent. The solutions are applied at rates of 25, 100, and 200 $\mu g/cm^2$ with a blade applicator. Strips treated with pyrethrins at 5 $\mu g/cm^2$ are used as standards for comparison.

In these tests, 8-in. strips of treated and untreated paper are joined edge-to-edge lengthwise with cellulose tape on the untreated side. Two such test surfaces are positioned so that the treated half of one is turned to the right and treated half of the other turned to the left to counteract any undetermined external influence on the distribution of the test insects. Two glass cylinders, 2.5 cm in height and 6.4 cm in inside diameter, are placed on each of the two sections of paper to provide test arenas of equal areas of treated and untreated paper. Two sets (four arenas each) of untreated paper matched with untreated paper are used as checks.

Ten confused flour beetle adults are exposed in each test arena, and the number of insects on the treated half and on the untreated half of the arena is recorded at 9 a.m. and 3 p.m. After application of the chemicals, exposures to determine the average numbers of insects on the untreated half of the repellency arena during a 5-day period is initiated at 4 days, 2 weeks, 1 month, and 2 months. The averages are converted to express "percent repellency or attractancy" by doubling the differences between the percentage of insects counted on the untreated half and the 50-percent distribution expected if only untreated papers are used. Positive figures (+) express repellency and negative figures (-) attractancy. They are listed as follows:

Class	Repellency (percent)
0	>-0.1 to <0.1
I	.1 to 20
II	20.1 to 40
III	40.1 to 60
IV	60.1 to 80
V	80.1 to 100

The same criteria are used for attractancy except that the repellency percentages are all negative. Class III is regarded as the minimum repellency for further consideration; however, the main selection factor is whether the chemical is more repellent than the pyrethrins-piperonyl butoxide standard or not.

Exhibit 7

TEST METHOD FOR INITIAL EVALUATION OF PROMISING INSECTICIDES AS PROTECTANTS FOR COMMODITIES

USDA-ARS, Stored-Product Insects Research and Development Laboratory
Savannah, Georgia

A. Formulation of Candidate Insecticides

1. Emulsifiable concentration as formulated by the manufacturer.

2. Or formulate technical as follows (wt./wt.).

a. Technical insecticide	25%
b. Emulsifier	10%
c. Xylene	65%

3. Or dust formulation as supplied by the manufacturer.

B. Rate of Application

1. 5 ppm (0.5 mg of actual insecticide per 100 grams of commodity)

2. 10 ppm (1.0 mg/100 gm)

3. 20 ppm (2.0 mg/100 gm)

(Check volume of spray to weight of commodity that will give good distribution and standardize accordingly. Note effect on moisture content.)

C. Method of Application

Kansas State University tumbler with 1-gal. jars for 15 minutes of tumbling. The subsamples are poured into the test jars directly from the treatment jar but the treated material is tumbled for a few seconds just prior to pouring each of the subsamples.

D. Test Commodities to be Used

Commodity	Moisture content	Temperature	Test Insects 1/
Wheat	12	80° F.	1, 2, + 3
Sorghum	14	80° F.	1 + 4

Commodity	Moisture content	Temperature	Test Insects 1/
Rice	12	80° F.	1, 2, 3, + 4
Corn	13	80° F.	1, 2, + 3
Peanuts	8	80° F.	1, 5 + 6
Blackeye cowpea	10	80° F.	7
Almonds (in shell)	10	80° F.	5 + 6
Natural raisins	16	80° F.	5 + 6

E. Test Containers

Treated commodity is to be placed in 1-qt. mason jars immediately after it is treated. Each jar should contain exactly 200 grams of a treated commodity. The opening of all containers is to be covered with 40-mesh wire cloth after insects are introduced.

F. Test Insects

Species	Stage	Age in days	No. in each container
1. Confused flour beetle	Adults	28 to 42	50
2. Rice weevil	Adults	28 to 42	50
3. Rice weevil	Larvae 2/	14 to 28	50
4. Lesser grain borer	Adults	14 to 21	50
5. Indian meal moth	Larvae	28	50
6. Indian meal moth	Eggs	2 to 4	50
7. Cowpea weevil	Adults	2 to 4	50

G. Replications

Five for each commodity, for each species, for each stage, for each type of test (initial toxicity and residual).

H. Exposure Period

1. Initial toxicity test

1/ See paragraph "F" for list of test insects.

2/ Internal infestaion.

a. Introduce insects 24 hours after treatment. Grain containing immature stages should be rolled with the treated grain in the test jars.

b. In those tests in which adults are used, remove insects using a U.S. Standard sieve No. 8 for wheat and No. 10 for corn after a 21-day exposure; record number dead, moribund, and alive immediately after separation and 24 hours later; discard insects; place samples back in original container with any dust that may have been removed during the separation of the insects and inspect again for presence of any form of insect life 42 days later and record as above. The separation of the insects shall be done using a rotary sifter and operated for 1 minute.

c. In those tests in which larvae and eggs are used, remove samples from container after a 56-day exposure, record all insects present, their stage and condition (dead, moribund, or alive). Separation of insects will be the same as specified above (H1b).

2. Residual life test

If initial toxicity test shows treatment to be effective, repeat procedure under H1 after treatment has aged 3, 6, 9, and 12 months. If initial toxicity test shows treatment to be ineffective, terminate test.

I. Temperature and Relative Humidity Requirements

All rearing of test insects, exposures, and postexposures shall be in rooms with a temperature of 80° F. and a relative humidity of 60±5%.

J. Standard and Untreated Checks

With each series of tests there will be a treated standard using malathion at 10 ppm and an untreated check.

References

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Exhibit 8

TEST FOR RESIDUAL TOXICITY AGAINST STORED-PRODUCT INSECTS

USDA-ARS, Stored-Product Insects Research and Development Laboratory
Savannah, Georgia

The insects used in the tests are adults of the confused flour beetle, *Tribolium confusum* Jacquelin duVal, 7 to 14 days old, and larvae of the black carpet beetle, *Attagenus megatoma* (F.), 3 to 5 months old.

The compounds are prepared in acetone solutions and applied to 3- by 12-in. strips of aluminum foil laminated to 40-lb. kraft paper with a Gardner automatic blade applicator. The compounds are first tried in an exploratory test as 1-day-old residues at rates of application of 50 $\mu g/cm^2$ on the aluminum surfaces and 100 $\mu g/cm^2$ on the paper surfaces. If a compound kills 50 percent or more of the insects on either surface, it is tested further as described below; however, if less than 50-percent kill is obtained on either surface, that surface is not tested further.

Four open-end glass cylinders, 6.4 cm in diameter and 2.5 cm in height, are placed on each treated surface. Ten confused flour beetles are placed in each cylinder and exposed 4 hours. Ten black carpet beetle larvae are placed in each cylinder on other treated strips and exposed 24 hours. The insects are then transferred to clean petri dishes for postexposure observations. The number of knocked down and dead-plus-moribund insects is recorded 120 hours after exposure for black carpet beetle larvae.

If the compound shows promise in the exploratory test, it is further tested by these same procedures at lower rates of 5, 10, and 50 $\mu g/cm^2$ on paper surfaces and 1, 5, and 25 $\mu g/cm^2$ on aluminum surfaces as 1-day-old residues. It is also tested as 28-day-old residues at rates of application of 10, 50, and 100 $\mu g/cm^2$ on paper surfaces and 5, 25, and 50 $\mu g/cm^2$ on aluminum surfaces. The flour beetles are exposed for 24 hours to the 28-day-old residues; otherwise, these tests are conducted the same as those for the 1-day-old residues. Malathion-treated surfaces are used as standards for comparison, and acetone-treated surfaces are used as controls.

Exhibit 9

STANDARD STAKE METHOD

USDA-Forest Service, Wood Products Insect Laboratory
Southern Forest Experiment Station
Gulfport, Miss.

1. Designed to simulate treatment of soil in trenches around foundation walls, piers, pilings, water and sewer lines, and other ground-to-building connections in crawl space and/or basement-type buildings.

2. In use since 1944 to present.

3. The original test unit design was to remove 2 cu ft (0.566 m^3) of soil to make 15 in. (38.1 cm) in diameter and 19 in. (48.3 cm) deep. This design was changed in 1956 by reducing the depth to 14-3/4 in. (37.4 cm) to give 1.5 cu ft (0.43 m^3). In both cases the rate of application of the chemical is the same, based on volume of soil treated; i.e., 4 gallons (18.2 liters)/10 cu ft (0.280 m^3). After the soil is treated with the insecticide in a water emulsion or oil solution and replaced in the hole, a 2- by 4- by 12-in. (5.08- by 10.16- by 31.0-cm) untreated pine sapwood stake is driven 6 in. (15.2 cm) deep into the center. The termites have to penetrate the treated soil to attack the stake.

4. Selection criteria of a site for a study should include the following:

a. Use land that has as little slope as possible to prevent sheet erosion of soil from one treatment to another.

b. Use land that has been preselected for a known termite population by a prebaiting survey or by history of the area.

c. Use land that is protected from fire and other disturbances and that will be available for at least 20 years.

5. Each treatment is replicated 10 times in a randomized block design with 5 ft (1.5 m) between each treatment, center to center. The stakes are carefully examined annually for termite attack. When 50 percent of the stakes of a treatment have been attacked, the treatment is considered to have failed. Stakes that are decayed are replaced at time of inspection.

6. Ten to thirty stakes within the study area are inserted in untreated soil to serve as checks.

Exhibit 10

STANDARD GROUND-BOARD METHOD

USDA-Forest Service, Wood Products Insect Laboratory
Southern Forest Experiment Station
Gulfport, Miss.

1. Designed to simulate treatment of the soil before pouring a concrete slab foundation.

2. In use since 1946 to present.

3. The original test unit design was a 2-ft (60.9-cm) square of soil from which all vegetation and duff were removed. In 1952 the area was reduced to a 17-in. (43.2-cm) square of soil. A known amount of chemical as a water emulsion or oil solution at 1 pint/sq ft (473 ml/ 929 cm^2) is spread over the soil surface. After the chemicals have soaked into the soil, an untreated sapwood pine board measuring 1 by 6 by 6 in. (2.5 by 15.2 by 15.2 cm) is laid flat in the center of the treated area. Termites have to penetrate the treated soil to attack the board.

4. Selection criteria of a site for a study should include the following:

a. Use land that has as little slope as possible to prevent sheet erosion of soil from one treatment to another.

b. Use land that has been preselected for a known termite population by a prebaiting survey or by history of the area.

c. Use land that is protected from fire and other disturbances and that will be available for at least 20 years.

5. Each treatment is replicated 10 times in a randomized block design with 5 ft. (1.5 m) between each treatment, center to center. The boards are carefully examined annually for termite attack. When 50 percent of the boards of a treatment have been attacked, the treatment is considered to have failed. Boards that are decayed are replaced at time of inspection.

6. Ten to thirty boards within the study area are placed on untreated soil to serve as checks.

7. When possible, the studies using this method are repeated in more than one location for comparison against different termite species in various soils and in varying climates.

Exhibit 11

MODIFIED GROUND-BOARD METHOD

USDA-Forest Service, Wood Products Insect Laboratory
Southern Forest Experiment Station
Gulfport, Miss.

1. Designed to more closely simulate the actual conditions underneath a concrete slab foundation which would exist in actual use than does the standard ground-board method.

2. In use since 1965 until present.

3. As in the standard ground-board method, all vegetation and duff are removed to expose the soil over a 24-in. (60.9-cm) square area. The chemicals are applied at 1 pint/sq ft (473 ml/929 cm^2) as water emulsions over an area 17 in. (43.2 cm) square in the middle of the cleared area. After the chemical soaks into the soil, a trench approximately 6 in. (15.2 cm) by 3.5 in. (8.9 cm) wide is excavated around the outside of the perimeter of the treated area. A polyethylene vapor barrier material is placed over the treated area and extends 1 inch (2.5 cm) into the trench on all sides. A short section, 4 in. (10.0 cm) diameter of plastic pipe is placed on end in the center of the area. Concrete is then poured over the treatment, around the pipe, and into the trench to form the simulated slab. When the concrete is set, the vapor barrier is removed from the area inside the pipe, and a 2- by 3- by 4-in. (5.8- by 7.6- by 10.0-cm) sapwood pine block is placed on the soil. A cap is placed over the end of the pipe to form a seal. Termites have to penetrate the treated soil to attack the block of wood.

4. Selection criteria of a site for a study should include the following:

a. Use land that has as little slope as possible to prevent sheet erosion of soil from one treatment to another.

b. Use land that has been preselected from a known termite population by a prebaiting survey or by history of the area.

c. Use land that is protected from fire and other disturbances and that will be available for at least 20 years.

5. Each treatment is replicated 10 times in a randomized block design with 5 ft. (1.5 m) between treatments, center to center. The wood blocks are carefully examined annually for termite attack. When 50 percent of the blocks of a treatment have been attacked, the treatment is considered to have failed. Blocks that are decayed are replaced at time of inspection.

6. Ten to twenty blocks within the study area are placed on untreated soil under cover to serve as checks.

7. When possible, the studies using this method are repeated in more than one location for comparison against different termite species, in various soils, and in varying climates.

Exhibit 12

STANDARD METHOD FOR LABORATORY EVALUATION TO DETERMINE RESISTANCE TO SUBTERRANEAN TERMITES (Standard M12-70)

American Wood Preservers Association
Washington, D.C.

1. Scope

1.1 This method provides for the evaluation of treated or untreated cellulosic material for its resistance to subterranean termites.

2. Apparatus and Material

2.1 Glass or clear plastic containers with loosely fitting tops, 240 ml (8 oz).

2.1.1 If volatile chemicals are to be tested, a 4.76-mm (3/16-in.) hold is drilled in the center of the top.

2.2 Screened, washed, heat-sterilized, brown or white sand.

2.3 Distilled water.

2.4 Plastic rings, 13 mm (1/2 in.) diameter by 6 mm (1/4 in.) long.

2.5 Southern yellow pine (*Pinus* spp.), 19 mm (3/4 in.) blocks, sapwood, no visisble defects, smoothed surfaces (planed or sanded) four to ten rings per 25 mm (1 in.).

2.5.1 Other wood species may be used, but in each separate test using other species as the major test wood, five southern yellow pine sapwood blocks must be used as additional controls to permit the correlation of test results among laboratories.

2.6 Subterranean termites. Using a major common species of the region in which the test is being run.

2.6.1 Specific identification of any termite used shall be obtained and reported with the test data.

2.7 Enamel or stainless steel tray, 254 by 508 mm (10 by 20 in.) and bucket.

2.8 Paper towels.

2.9 Zephiran chloride solution (1 part zephiran chloride to 750 parts water)

3. Determination of Sand Water-Holding Capacity

3.1 The quantity of distilled water to be added to the sand during the test shall be determined as follows:

3.1.1 Place 100 g of oven-dry sand in a beaker and determine the volume of water required to saturate the sand. The saturation point is defined as the point when the addition of more water will result in free water on the surface of the sand.

3.1.2 Calculate the percent saturation as follows:

$$\text{Percent saturation} = \frac{\text{Weight water}}{\text{Weight sand + Weight water}} \times 100$$

3.1.3 Add water to the sand as follows:

Percent water to add = Percent saturation - 5.0

3.1.4 As an example:

Saturation point was reached at 20 ml of water.

$$\text{Percent saturation} = \frac{20}{120} \times 100 = 16.7 \text{ percent.}$$

Percent water to add = 16.7 - 5.0 = 11.7 percent.

4. Collection of Termites

4.1 Subterranean termites (*Reticulitermes, Coptotermes,* etc., spp.) are collected from a natural forest situation; e.g., from fallen logs or from stumps.

4.1.1 Short log sections are removed to the laboratory and are split. The insects are shaken out onto a tray or trays. After distributing the debris and insects evenly on the tray(s), damp paper towels are laid over the debris. The termites will cling to the damp paper after a few minutes.

4.1.2 A 7.57- to 11.35-liter (2- to 3-gal) pail is prepared by placing about 10 unfolded slightly crumpled damp paper towels in the bottom of the pail. These towels should be rinsed in distilled water and squeezed damp a number of times. Cover these towels with about 10 unfolded dry paper towels.

4.1.3 The damp towels covering the tray debris are shaken into the above described pail. After 2 to 4 hr the dry towels and any insects and debris on them are removed from the pail and discarded. Insects clinging to the lower damp towels are used in the test.

4.1.4 Termites should not be held in the pail longer than 24 hr before being used.

4.1.5 CAUTION: Exercise reasonable care to insure that any termites discarded (4.1.3) are dead. Oven-drying in the debris and towels used at 100° C for 6 hr is sufficient. When a test is finished, reasonable care should be exercised that living insects are not discarded.

5. Weathering the Test Blocks

5.1 If the test material is weathered prior to exposure to the insects, the complete details on the weathering shall be reported.

5.2 The ASTM weathering procedure for the soil block test is recommended (ASTM D 1413-61).

6. Conditioning of the Test Blocks

6.1 All test blocks, following weathering if used, shall be conditioned to a constant weight and the individual weights recorded, prior to exposure to the insects.

6.2 The ASTM soil block conditioning procedure is recommended (ASTM 1413-61).

7. Block Quantity and Identification

7.1 Five replicate blocks should be prepared for each variable under test; e.g., for each retention of each preservative or chemical to be tested.

7.2 Five untreated blocks of the same species as the blocks in 7.1 must be used as controls for each separate study.

7.3 If southern yellow pine (SYP) is not used as the species in 7.1 and 7.2, then five blocks of untreated SYP must be added to each study to permit a comparison to studies using SYP as the major species.

7.4 All blocks must be identified with a number in a suitable manner.

8. Assembling Containers

8.1 Prior to using, all containers (2.1) shall be washed, rinsed in the Zephiran chloride solution, and dried.

8.2 Sand in amount of 250 g is added in one or two increments to each container.

8.2.1 If a material which is easily leachable from wood is under test, all the sand is added to the container at one time and the individual weighed block is placed on a plastic ring on the surface of the sand in the center of the container. This will keep the block slightly above the substrate.

8.2.2 If the material under test is not readily leachable, the sand is added in two increments to each container as follows:

8.2.2.1 Fill each container about one-half with sand and press one of the weighted test blocks about half way into the sand with one side of the block touching the container side.

8.2.2.2 Add the second increment of sand, filling each container about two-thirds full.

8.2.3 Five containers are prepared without blocks but with the same quantity of sand as above. These containers are used to determine the natural vigor of the insects used in the study.

8.3 Mark or number each container for identification.

8.4 Sufficient distilled water is added to each container as determined in Section 3. After addition of the water, the containers are set aside for 24 hr.

9. Adding Termites

9.1 One-half g (±0.01 g) of subterranean termites (Section 4) are weighed and added to each of the previously prepared containers.

9.2 The container tops are replaced loosely.

10. Container Storage and Inspections

10.1 The test containers are weighed individually and maintained at 25 to 28° C. for 30 days.

10.1.1 Every 5 to 7 days the containers are examined and the presence of tunneling, termite mortality, and position of the termites in the container recorded as follows:

10.1.1.1 Tunnelling present - yes, no.

10.1.1.2 Majority termite position - up, down.

10.1.1.3 Termite mortality - none, slight, moderate, heavy.

10.2 Periodically weigh five randomly selected containers and add distilled water if the moisture content of the sand drops below two percentage points of the original moisture content (see Section 3).

11. Container Disassembly

11.1 After 30 days, the containers are disassembled and the blocks removed and cleaned. Prior to and during the disassembly the items in Section 10.1.1 are noted.

12. Block Evaluation

12.1 Each block is examined and visually rated using the termite rating system as given in "Standard Method of Evaluating Wood Preservatives by Field Tests with Stakes," ASTM D 1758-62, as follows:

Recording Grade	Numerical Rating	Description of Condition
A	10	Sound
B	9	Trace of attack
C	7	Moderate attack
D	4	Heavy attack
E	0	Failure by termite attack

Two rating scales are shown for termite rating. This is done to prevent confusion when recording data. When analyzing the ratings, the numerical ones can be substituted. If desired, the numerical ratings may be used throughout.

12.2 Following the above rating, the blocks are reconditioned to constant weight under the same conditions used in 6.1.

12.2.1 The individual block weight losses are determined.

12.3 The visual and weight loss ratings are correlated for each block and each group of replicates. A single combined rating may be assigned to each block if desired.

12.4 A statistical analysis of the data should be completed and a treatment evaluation derived.

Exhibit 13

TEST METHOD FOR ANTIFOULING PAINTS ON WOOD SUBSTRATES FOR FRESH OR SALT WATER EXPOSURE

Adapted from Environmental Protection Agency and Industry Sources

1. Scope

1.1 This procedure outlines the testing procedure of marine antifouling paints for wood substrates. It directs how panels are to be prepared and exposed for testing to acquire efficacy information to be submitted with application for registration.

2. Test Panel Preparation

2.1 The panels may be prepared either in the laboratory or at the exposure site.

2.2 Recommended test panels are 13 mm (1/2 in.) thick Douglas Fir plywood, exterior grade or Marine, solid core, and at least 152 by 304 mm (6 by 12 in.)

2.3 Prime and paint panels as directed on the label of the paint being tested.

2.4 Exposed at each test location at least two panels for each paint to be tested, two standard panels painted with three coats U.S.N. Formula 121/63; and two check (control) panels painted with a brown alkyd enamel without toxicant.

2.5 Prepare all wood panels as follows:

2.5.1 Fill all core openings or defects in panel stock with solvent-type wood filler. After filler has dried, sand all surfaces.

2.5.2 Apply test paint as directed on label.

2.5.3 Prepare standard 121/63 panels as follows:

2.5.3.1 Apply two coats of U.S. Navy Formula 121/63 (MIL-P-15931B) with brush, roller or spray gun to a dry-film thickness of 4.0 mils (minimum). Allow minimum of 1 hr drying between coats (minimum 21° C and 70% or less R.H.). Allow minimum of 4 hr drying of last coat before immersion.

2.5.4 Prepare check (control) panels as follows:

2.5.4.1 Apply two coats of brown alkyd enamel approximating Federal Standard Color #30166. Apply to a dry film thickness of 4.0 mils minimum. Refer to ASTM D2691-70 Dry Film Thickness of Coating on Wood Products. Allow at least 24 hours between coats and between last coat and immersion. Enamel used shall conform with Federal Specification TT-E-490B. The color specified approximates the color of 121/63.

2.6 Shipment shall be made in either slotted boxes, to keep panels separate, or in polyethylene sleeves, each panel being sealed in a separate pocket. Panels shall be shipped in fresh or sea-water (depending upon exposure) when labeling calls for specified launching time after painting.

3. Exposure Method

3.1 Sample panels may be exposed in any standard racks available at each exposure station. Local conditions may influence exposure depth.

3.1.1 For totally submerged panels, it is recommended that they be suspended from either a float or a fixed support so that top of panels are at least 1 foot below water surface.

3.2 All samples from one shipment should be exposed on the same day.

4. Examination

4.1 Panels shall be examined and reported on at 30-day intervals.

4.2 From each examination a Fouling Resistance (F.R.) statistic will be developed, the panels being rated as follows:

	Score
Surface free of attached fouling organism	100
Incipient forms present only	95

If mature forms are present, subtract from 100 or 95 the total number of individual organisms present, or the total percent area covered by colonial forms.

Example

		Score
No incipient fouling, panel clean		100
Barnales -10 each	-10	
Tunicates -2 each	-2	
Algae -15 each	-15	

Example

Score
100
-27

F.R. (Fouling Resistance) = 73

4.3 Each panel of the set is graded as above, as are the standards and the untreated checks.

4.4 Identification of fouling organisms to genus is not necessary. However, if such identification to genus and species is possible, so report, identifying types of organism (Barnacle,Tunicate, Algae, etc.).

4.5 Grade paint film as follows:

4.5.1 A film with no defects scores 100.

4.5.2 Subtract the percentage of the surface showing defects from 100.

4.5.3 This statistic is called A.F. (Antifouling film rating).

5. Reporting

5.1 Efficacy for supporting application shall be based on 12 months' exposure.

5.2 Copies of each monthly report shall be submitted with application.

5.2.1 Reports should show following information:

1. Identity of exposure station and location
2. Type of exposure
3. Length of exposure with dates
4. Identity of paints
5. Fouling resistance rating for each panel
6. Antifouling film rating for each panel

Exhibit 14

TEST METHOD FOR ANTIFOULING PAINTS ON METAL SUBSTRATES
FOR FRESH OR SALT WATER EXPOSURE

Adapted from Environmental Protection Agency and
Industry Sources

1. Scope

1.1 This method describes the preparation and testing of fresh and salt-water antifouling paints on steel and aluminum substrates.

2. Test Panels

2.1 All panels should be prepared as follows unless these directions conflict with label directions of the paint being tested. In any case, standard and control panels should be prepared as follows:

2.2 Steel panels

2.2.1 The recommended panel is a medium (mild) steel plate (low carbon, MIL-S-22698, Type 3, Class A or as covered by ASTM A569), at least 3 by 152 by 305 mm (1/8 by 6 by 12 in.), with a minimum of 466 square cm (72 square in.) per side. A 6-mm (1/4-in.) diamter hole, 6 mm from the top and centered, shall be drilled for holding the panel for handling and while painting. A 19-mm (3/4-in.) vinyl tape numbered and applied between the first and second coats of antifoulding paint can be used for identification.

2.2.2 Surface preparation: The panel should be abrasive blasted to near white metal.

2.2.3 Coating system used will depend on paint being tested. All steel panels should be stored in a heated drying oven 82° C immediately after blasting if the panels are not to be coated immediately. Such treatment will prevent rust on the blasted surface.

2.2.4 Standard panel preparation - Antifouling.

2.2.4.1 One coat (0.5 mil or 13 microns) #117 pretreatment coating, MIL-P-15328.

2.2.4.2 Four coats (6.0 mils or 150 microns) #119 Vinyl Red Lead Primer, MIL-P-15929.

2.2.4.3 Two coats (4.0 mils or 100 microns) #121/63 Red Vinyl (A.F.), MIL-P-15931.

2.2.5 Drying times:

2.2.5.1 The abrasive blasted panel should be coated with the #117 within 8 hr after blasting.

2.2.5.2 The #117 should be top coated with the first coat of #119 red lead vinyl within 24 hr.

2.2.5.3 Allow a minimum of 1 hr drying time between the next coats of #119 and 121/63 (minimum 21° C and relative humidity of 70% or less) and a maximum of 24 hr. Allow a minimum of 4 hr drying for the last coat of 121/63 A.F. before immersion and a maximum of 2 weeks (this allows shipping time of the panel to the immersion site).

2.2.6 Primer will be used as recommended on label or accompanying literature, i.e., Technical Bulletins, etc.

2.3 Aluminum panels

2.3.1 Aluminum panels shall conform both in substance and preparation to ASTM D1733-63 "Standard Method of Preparation of Aluminum Alloy Panels for Testing Paint, Varnish, Lacquer, and Related Products."

2.3.2 Proceed with the coating system as specified on label of paint on test.

2.4 Expose at each test location at least two panels for each paint to be tested; two panels painted with the standard paint system and exposed with suitable uncoated blacks (slate panels may be used as satisfactory black surface).

2.5 Shipment should be made in either slotted boxes, to keep panels separate, or in polyethylene sleeves, each panel being sealed in a separate pocket. Panels should be shipped in fresh or sea-water (depending upon exposure) when labeling calls for specified maximum launching time after painting.

3. Exposure Method

3.1 Sample panels may be exposed in any standard racks available at each exposure station. Local conditions may influence depth.

3.1.1 For totally submerged panels, it is recommended that they be suspended from either a float or a fixed support so that top of panels are at least 1 foot below water surface.

3.2 All samples from one shipment must be exposed on the same day.

4. Examination

4.1 Panels should be examined and reported on at 30-day intervals.

4.2 From each examination a Fouling Resistance (F.R.) statistic will be developed, the panels being rated as follows:

	Score
Surface free of attached fouling organism	100
Incipient forms present only	95
If mature forms are present, subtract from 100 or 95 the total number of individual organisms present, or the toal percent area covered by colonial forms	

Example

		Score
No incipient fouling, panel clean		100
Barnacles -10 each	-10	
Tunicates -2 each	-2	
Algae -15 each	-15	
		-27
F.R. (Fouling Resistance) =		73

4.3 Each panel of the set is graded as above, as are the standards and the untreated checks.

4.4 Identification of fouling organisms to genus is not necessary. However, if such identification to genus and species is possible, so report, identifying type of organism (Barnacle, Tunicate, Algae, etc.).

4.5 Grade paint film as follows:

4.5.1 A film with no defects scores 100.

4.5.2 Subtract the percentage of the surface showing defects from 100.

4.5.3 This statistic is called A.F. (Antifouling film rating).

Exhibit 15

TEMPORARY FABRIC TREATMENTS

USDA-ARS, Stored Product Insects Research and Development Laboratory
Savannah, Ga.

1. Species

Clothes moth and carpet beetles

Webbing clothes moth, *Tineola bisselliella* (Hummel), and the black carpet beetle, *Attagenus megatoma* (F.), are preferred. Both larvae and adults should be tested.

2. Number of test insects

Four sets of 10 insects replicated 3 times (120 insects of each stage).

3. Observations

Insects should be observed 24 and 48 hours after treatment, and the percentage of knocked-down (KD) and dead-plus-moribund (D+M) insects recorded. Data should be presented in tabular form.

NOTE: Some nonresidual sprays may also function as short-term protectants or semipermanent mothproofers when sprayed directly on woolen fabric. If classification as a semipermanent mothproofer is desired, the criteria in the following section on semipermanent mothproofers should be followed.

Reference

Bry, R.E., J.H. Lang, and R.E. Boatright. 1973. Toxicity of resmethrin to carpet beetles and clothes moths. *Pest Control* 41 (11): 32, 47.

Exhibit 16

SEMIPERMANENT FABRIC TREATMENTS

USDA-ARS, Stored Product Insects Research and Development Laboratory
Savannah, Ga.

1. Application and aging

Official moth test cloth should be sprayed until "damp" to the touch. Insect-feeding tests should be conducted when the cloth is dry. Samples should be stored under darkened or semidarkened conditions, and feeding tests should be conducted at selected intervals during the aging period. The efficacy date submitted must show that the treatment satisfactorily protects the cloth for the time period claimed. Chemical analyses should be conducted initially and at each aging interval.

2. Biological test procedures

Use the applicable procedures as published by the Chemical Specialties Manufacturers Association (CSMA) or the American Association of Textile Chemists and Colorists (AATCC).

3. Species

Clothes moths and carpet beetles

Webbing clothes moth, *Tineola bisselliella* (Hummel), and black beetle *Attagenus megatoma* (F.), are preferred.

4. Observations

The appropriate excrement-weight or fabric-weight-loss data, mortality data and the analytical results should be submitted in tabular form. Visual feeding damage ratings are required.

NOTE: If the semipermanent mothproofer is applied in some other manner such as from a drycleaning solution or from a padding machine, the above aging, biological test procedures, and manner of reporting will apply.

Reference

Bry, R.E., R.E. Boatright, J.H. Lang, and R.S. Cail. 1973. Protecting woolen fabric against insect damage with resmethrin. *Soap Cosm. Chem. Spec.* 49(3): 40, 42, 44.

Exhibit 17

PERMANENT FABRIC TREATMENTS

USDA-ARS, Stored Product Insects Research and Development Laboratory
Savannah, Ga.

1. Laboratory application procedures

Permanent mothproofers are usually applied to woolen fabric during dyeing, and efficacy data should generally be generated with a mill run; however, laboratory procedures such as that described by Bry and Simonaitis (1975) will be a close simulation of the dyebath process.

2. Performance test procedures

Performance test procedures developed by AATCC (AATCC Test Method 28-1974--same as American National Standards Institute L 14-65-1960/R1971) should be employed. These criteria should be considered as the minimum requirements for a permanent mothproofer and additional extended testing should be conducted. The following table lists the minimum requirements and the suggested additional testing

Manipulation	Minimum requirements	Maximum requirements
Washing*	5 times	10 - 15 times
Drycleaning:		
Stoddard solvent or perchlorbethylene	5 times	10 - 15 times
Hot pressing	5 times	10 - 15 times
Sea water	5 immersions	
Perspiration:		
Acid	5 immersions	
Alkaline	5 immersions	
Light:		
Fade-Ometer	40 Standard Fading Hours	100 Standard Fading Hours

NOTE: Chemical analyses should be conducted before and after the above manipulations.

*Procedure described by Bry and Lang (1967) may be substituted.

3. Biological test procedures

Use the applicable procedure published by CSMA or AATCC (AATCC Test Method 24-1974).

4. Species

Clothes moths and carpet beetles

Webbing clothes moth, *Tineola bisselliella* (Hummel), and black carpet beetle, *Attagenus megatoma* (F.), are preferred.

5. Observations

The appropriate excrement-weight or fabric-weight-loss data, mortality, and the analytical results should be submitted in tabular form. Visual feeding damage ratings are required.

References

Bry, R.E., and J.H. Lang. 1967. 0,0-diethyl phosporothioate O-ester with phenylglyoxylonitrile oxime (Bay 77488) as a mothproofer of woolen fabric. *Textile Res. J.* 37(11): 915-919.

Bry, R. E., and R.A. Simonaitis. 1975. Mothproofing in an acid dyebath. *Textile Chem. Color.* 7(2): 28-29.